



THE UNIVERSITY *of* EDINBURGH

Edinburgh Research Explorer

Sexual dimorphism of the tibia in contemporary Greek-Cypriots and Cretans

Citation for published version:

Kranioti, EF, García-Donas, JG, Almeida, PSP, Kyriakou, X-P & Langstaff, HK 2017, 'Sexual dimorphism of the tibia in contemporary Greek-Cypriots and Cretans: Forensic applications' *Forensic Science International*, vol. 271. DOI: 10.1016/j.forsciint.2016.11.018

Digital Object Identifier (DOI):

[10.1016/j.forsciint.2016.11.018](https://doi.org/10.1016/j.forsciint.2016.11.018)

Link:

[Link to publication record in Edinburgh Research Explorer](#)

Document Version:

Peer reviewed version

Published In:

Forensic Science International

General rights

Copyright for the publications made accessible via the Edinburgh Research Explorer is retained by the author(s) and / or other copyright owners and it is a condition of accessing these publications that users recognise and abide by the legal requirements associated with these rights.

Take down policy

The University of Edinburgh has made every reasonable effort to ensure that Edinburgh Research Explorer content complies with UK legislation. If you believe that the public display of this file breaches copyright please contact openaccess@ed.ac.uk providing details, and we will remove access to the work immediately and investigate your claim.



Accepted Manuscript

Title: Sexual dimorphism of the tibia in contemporary Greek-Cypriots and Cretans: forensic applications.

Author: Kranioti EF García-Donas JG Prado Almeida PS
Xenia-Paula Kyriakou Langstaff HK



PII: S0379-0738(16)30494-7
DOI: <http://dx.doi.org/doi:10.1016/j.forsciint.2016.11.018>
Reference: FSI 8652

To appear in: *FSI*

Received date: 3-6-2016
Revised date: 5-11-2016
Accepted date: 10-11-2016

Please cite this article as: EF Kranioti, JG García-Donas, PS Prado Almeida, Xenia-Paula Kyriakou, HK Langstaff, Sexual dimorphism of the tibia in contemporary Greek-Cypriots and Cretans: forensic applications., Forensic Science International <http://dx.doi.org/10.1016/j.forsciint.2016.11.018>

This is a PDF file of an unedited manuscript that has been accepted for publication. As a service to our customers we are providing this early version of the manuscript. The manuscript will undergo copyediting, typesetting, and review of the resulting proof before it is published in its final form. Please note that during the production process errors may be discovered which could affect the content, and all legal disclaimers that apply to the journal pertain.

Sexual dimorphism of the tibia in contemporary Greek-Cypriots and Cretans: forensic applications.

Kranioti EF^{1,2}, García-Donas JG¹, Almeida Prado PS^{3,4}, Xenia-Paula Kyriakou⁵, Langstaff HK¹

¹Edinburgh Unit for Forensic Anthropology, School of History Classics and Archaeology, University of Edinburgh, 4 Teviot place, EH8 9AG, Edinburgh, UK.

²Department of Forensic Sciences, University of Crete, Medical School, Heraklion, Greece.

³Department of Biomorphology, Federal University of Bahia, Av. Reitor Miguel Calmon, s/n, Salvador/BA, 40110-100, Brazil.

⁴Medical Legal Institute Nina Rodrigues, Departamento de Polícia Técnica do Estado da Bahia, Av. Centenário s/n, Salvador/BA, 40.100-180, Brazil.

⁵Department of Bioarchaeology, Institute of Archaeology, University of Warsaw, ul. Krakowskie Przedmieście 26/28, 00-927 Warsaw, Poland.

Author for correspondence and reprint requests:

Elena F. Kranioti

Edinburgh Unit for Forensic Anthropology,

School of History, Classics and Archaeology, University of Edinburgh

William Robertson Wing, Old Medical School, Teviot Place,

Edinburgh, EH8 9AG

Tel. +44 (0)131 650 2368

Fax. +44 (0)131 650 2378,

E-mail: elena.kranioti@ed.ac.uk

Abstract:

Sex estimation is an essential step in the identification process of unknown heavily decomposed human remains as it eliminates all possible matches of the opposite sex from the missing person's database. Osteometric methods constitute a reliable approach for sex estimation and considering the variation of sexual dimorphism between and within populations; standards for specific populations are required to ensure accurate results. The current study aspires to contribute osteometric data on the tibia from contemporary Greek-Cypriots to assist the identification process. A secondary goal involves osteometric comparison with data from Crete, a Greek island with similar cultural and dietary customs and environmental conditions. Left tibiae from one hundred and thirty-two skeletons (70 males and 62 females) of Greek-Cypriots and one hundred and fifty-seven skeletons (85 males, 72 females) of Cretans were measured. Seven standard metric variables including Maximum length (ML), Upper epiphyseal breadth (UB), Nutrient foramen anteroposterior diameter (NFap), Nutrient Foramen transverse diameter (NFtrsv), Nutrient foramen circumference (NFCirc), Minimum circumference (MinCirc) and Lower epiphyseal breadth (LB) were compared between sexes and populations. Univariate and multivariate discriminant functions were developed and posterior probabilities were calculated for each sample. Results confirmed the existence of sexual dimorphism of the tibia in both samples as well as the pooled sample. Classification accuracy for univariate functions ranged from 78% to 85% for Greek-Cypriots and from 69% to 83% for Cretans. The best multivariate equations after cross-validation resulted in 87% for Greek-Cypriots and 90% accuracy for Cretans. When the samples were pooled accuracy reached 87% with over 95% confidence for about one third of the population. Estimates with over 95% of posterior probability can be considered reliable while any less than 80% should be treated with caution. This work constitutes the initial step towards the creation of an osteometric database for Greek-Cypriots and we hope it can contribute to the biological profiling and identification of the missing and to potential forensic cases of unknown skeletal remains both in Cyprus and Crete.

Key words: Greek-Cypriots, Cretans, Tibia, Sex estimation, Discriminant Function Analysis, Posterior Probabilities.

Introduction:

Since the summer of 1974 when Turkey invaded Cyprus and placed a large part of the territory of the Republic of Cyprus under its control the fate of hundreds of people remains unknown. A total of 1508 Greek-Cypriots and 493 Turkish-Cypriots were declared missing during that period [1], and despite the efforts of the Committee of Missing Persons [2] in Cyprus only 629 were identified and returned to their families. Identification mostly relied on DNA analysis or comparison of antemortem dental records. Yet, for these methods to be applied, one must have an indication as to the biological profile of the individual in order to narrow down the possible matches from the missing person's database. Thus, osteometric standards from contemporary skeletons of both Turkish-Cypriots and Greek-Cypriots are essential.

Macroscopic observation of the pelvis and/or the skull for sex estimation have been carried out by many researchers both in physical and virtual bones producing accuracy rates over 85% for both skeletal elements [e.g. 3-6]. The pelvic bone is the most reliable single sex indicator in the human skeleton exhibiting in certain occasions over 95% correct sexual diagnosis [4]. This can be attributed to the fact that sexual dimorphism of this skeletal element is related to functional adaptation of the female pelvis to support childbirth and thus it is population-independent. As an alternative to the traditional macroscopic examination, metric techniques allow accurate sexing through statistical analysis and therefore they provide a more rigorous assessment than the morphological approach [7]. Moreover, osteometric methods are long acknowledged to be valid for sex estimation of unknown skeletal remains when population-specific standards are employed [e.g. 4, 8-9]. Long bones were disregarded in the past due to the belief that skull is the second most dimorphic bone of the skeleton after the pelvis. Recent data support that long bones actually perform well for sex estimation reporting high classification accuracies [e.g. 9-11]. Amongst them, tibia was extensively studied in populations of different chronology and geography [12-21] being robust, likely to survive harsh taphonomic conditions and scavenging in decomposed bodies found outdoors. Thus, many studies have tested different measurements on the tibia in different populations with sex allocation accuracy reaching 98% in some cases [e.g. 16]. Sexual dimorphism of the tibia during ontogeny was also reported in a European sample [22]. These results imply that tibia is extremely dimorphic and thus an element with great discriminatory power.

In addition to the traditional osteometric studies which routinely use discriminant function analysis to create sex estimation formulae, new methodologies on data acquisition and analysis have been emerging recently [23-24]. Three-dimensional reconstructions of bones from CT scans or surface scans allowed for virtual measurements in every bone of the skeleton and the

development of sex estimation methods [25-26]. Furthermore, methods on quantifying size and shape such as geometric morphometrics [27-28] and machine learning approaches [29-30] also emerged. The possibility of creating osteological digital databases with access for researchers all over the world seems to have given an extra push in virtual methods. This led to creating population specific standards even in countries and regions lacking documented skeletal collections.

To date there are no data available on modern Greek-Cypriots while a large number of studies have been emerging recently for neighbouring Cretans [9,21, 31-35] and mainland Greeks [36-37]. The same situation holds true for Turkish-Cypriots with an increasing number of studies from mainland Turkey appearing recently [26, 38-41]. The main objective of this work is to develop population specific osteometric standards of the tibia for the Greek-Cypriots in an effort to contribute to the existing sex estimation methods employed for the identification of the missing in Cyprus. A secondary objective is to compare the dimensions of the tibia of two synchronous cemetery populations from Cyprus and Crete that share similar language, culture, dietary habits and climate.

Material and Methods:

One hundred and thirty-two skeletons (70 males and 62 females) were selected at random from a cemetery population housed in the ossuary of the main cemetery in the city of Limassol in Cyprus. The sample consisted of individuals who died between 1976 and 2003. The mean age for males was 69.3 ± 12 years and 70 ± 17.8 years for females. A contemporary collection from Crete, Greece was used for comparison [9]. One hundred and fifty-seven skeletons (85 males, 72 females) with mean age 68.8 ± 14 years for males and 70.9 ± 17.8 years for females were used in this study. Seven measurements [4, 12] were taken on the left tibia: Maximum length (ML), Upper epiphyseal breadth (UB), Nutrient foramen anteroposterior diameter (NFap), Nutrient Foramen transverse diameter (NFtrsv), Nutrient foramen circumference (NFCirc), Minimum circumference (MinCirc) and Lower epiphyseal breadth (LB).

Technical measurement error (TEM) was used to assess intra-observer error in a sample of 30 randomly selected bones. The relative TEM (rTEM), which expresses the error as a percentage of TEM divided by the average value for each measurement was also taken in order to scale the error. The coefficient of reliability (R) of the measurement is also calculated as suggested by Ulijaszek and Kerr [42].

The mean differences of the measurements between the population samples were tested using an independent T-test. Sex differences on the measurements were explored using a one-way

ANOVA. Additionally, univariate and multivariate discriminant functions were developed for the Greek-Cypriot (Cy), the Cretan (Cr) and the pooled (P) sample. Multivariate equations were created using different combinations of variables for each sample. The functions FCy1, FCr1 and FP1 used all available variables with a direct procedure while FCy2, FCr2 and FP2 are the result of stepwise discriminant function analysis. In addition, three more equations were created for each sample in an effort to simulate different fragmented scenarios. FCy3, FCr3 and FP3 employ the four variables of the upper epiphysis (UB, NFap, NFtrsv and NFCirc) allowing for the rest of the bone to not be available. For cases where the upper epiphysis is missing while the lower part is preserved FCy4, FCr4 and FP4 were created using LB and MinCirc. Lastly, in the case both epiphyses are missing FCy5, FCr5 and FP5 provide a method of sex classification by using only the three measurements at the nutrient foramen (NFap, NFtrsv, NFCirc). In this last scenario MinCirc was omitted as it would be very difficult to define the measurement in a bone missing the lower epiphysis.

Posterior probabilities were calculated for the best equations as described in Kranioti & Apostol [21]. Statistical analysis was carried out with SPSS 22.0.

Results:

Intra-observer error

Thirty randomly selected tibiae were measured by the same observer within 4 weeks of the first measurement. TEM, rTEM and R for each variable are presented in Table 2. rTEM is below 5% in all cases while R is consistently over 0.95 with the exception of TLB which is slightly lower. This is in accordance with the acceptable human error (rTEM<5%, R>0.95) as suggested by Ulijaszek and Kerr [42].

Inter-observer error

Fifteen randomly selected tibiae were measured by two independent observers. TEM, rTEM and R for each variable are presented in Table 2. rTEM is below 5% in all cases while R ranged between 0.69 (NFtrsv) and 0.96 (ML).

Sexual dimorphism

A Shapiro-Wilk's test ($p < 0.05$) and a visual inspection of the histograms, Q-Q plots and box plots were used to assess normal distribution. In some occasions data were not normally distributed for females in both sexes. Yet, in large samples ($N > 40$) the violation of normality is not expected

to bias the results [43-44]. A bootstrapping approach was used in the analysis since the sampling distribution tends to be normal independently of the shape of the data [43]. Bootstrapping confirmed that 1000 subsamples presented nearly normal distribution. ANOVA and Wilcoxon test confirmed the mean differences between the two sexes ($p < 0.001$) for all variables in both samples. F-values are generally higher for Cretans compared to Greek-Cypriots. More specifically the highest F-value is marked for NFap for Cretans and LB for Greek-Cypriots. Mean values, standard deviations and F-values are illustrated in Table 2.

Population differences

The whole sample was divided into males and females and mean differences for each subgroup were compared using an independent t-test using bootstrapping. Again, in several occasions data were not found to be normally distributed but the large sample overcomes the problem [43-44]. Two variables (ML, NFtrsv) were found to differ significantly ($p < 0.05$) between Cretans and Greek-Cypriots for both males and females. The mean ML value was more than 11mm greater for males and more than 9 mm greater for females from Cyprus, compared to the subgroups from Crete. In contrast, NFtrsv is consistently lower in mean value for Cypriots compared to Cretans for both sexes. Table 3 illustrates means and standard deviations for each subgroup, and t-values for each comparison. In addition, a Wilcoxon two-tailed test with a Monte-Carlo simulation of 1000 iterations was also done resulting in statistically significant differences between females for LB ($p < 0.027$).

Discriminant functions

Classification accuracy for univariate functions ranged from 78% to 85% for Greek-Cypriots, from 69% to 83% for Cretans and from 73% to 84% for the pooled sample. The best single variable was UB for Greek-Cypriots (85.2%) and the pooled sample (83.5%) and LB for Cretans (82.7%). Demarking points and classification accuracy for original and cross-validated data can be found in Table 4.

The most accurate multivariate equation for Greek-Cypriots was Cy4 (87%) which uses two variables (LB, MinCirc) and classifies 16% of the sample with >95% posterior probability of correct classification. Cy1 gives the second best overall accuracy with, however, a percentage of 29% of the sample to be correctly classified with >95% posterior probability of correct classification. For Cretans the best equations were FCr1 and FCr2 with classification accuracy of 90% for both original and cross-validated data. FCr2 is preferable due to the fact that it uses less variables and classifies >44% of the sample with >95% probability of correct classification. Table 5 illustrates all multivariate discriminant functions, classification accuracy for both

original and cross-validated data and the percentage of correct classification with >95% posterior probability. For the pooled sample FP2 (ML, NFap, LB) is the formula with the higher accuracy (87.1%) with about one third of the sample to be accurately classified with over 95% posterior probability. As a general remark the vast majority of the formulae performed better for males compared to females.

Discriminant functions were also calculated for males and females with the objective of exploring whether the ethnicity could be estimated. For males a multivariate equation using 3 variables (ML, UB and NFtrsv) resulted in 67% correct classification of Cretans and 74% of Greek-Cypriots. Similarly, an equation based on the same variables resulted in 74% of correct classification for Cretans females and 76% of Greek-Cypriot females. Again bootstrapping was used in this analysis.

Discussion:

Methods employed in forensic casework should follow the existing evidentiary rules (e.g. Daubert standards) for the admissibility of scientific evidence; hence reliability and accuracy must be calculated and error rates must be reported. In the same vein the Criminal Practice Directions Amendment No.2 [2014] EWCA Crim. 1569 (at paragraph V33A.5-6) in the Guidance on Expert Evidence by the Crown Prosecution Service in the UK highlights the importance of the reliability of the method in the admissibility of evidence. The recommendations for good practise in forensic anthropology by the American Board of Forensic Anthropology [45], the British Association for Forensic Anthropology [COP BAHID, 2015] [46] and the Forensic Anthropology Society Europe [FASE Basic Workshop, Coimbra, 2016] [47] mandate the application of population specific standards for any method of biological profiling; thus this work constitutes a step in the development of osteometric standards for Cyprus. Biometric data for Cretans have been published before for cranial and postcranial elements [9, 21, 31-35] including a study on the tibia [21] which however used only 3 of the 7 variables analysed herein.

Tibia has been extensively studied and used for the development of sex estimation methods due to its robusticity and its resistance to taphonomic agents [48]. Studies on the tibia on different populations employing different combinations of tibia measurements are presented in Table 6. For example, Slaus and Tomicic' [49] used six variables from the tibia in a Croatian sample providing accuracy of 92%. Lower accuracy rates were achieved by Işcan and Miller-Shaivitz [50] who found that tibia nutrient foramen provided an accuracy of 80% in whites and black individuals from the Terry Collection emphasizing the need of population specific standards. Gonzalez-Reimersa and colleagues [51] carried out a metric study on a prehispanic Canarian

population obtaining promising results (98% of correct sex classification) using a single variable. The authors argued that this parameter –tibia breadth- is useful for sexual dimorphism because it maximises muscular development differences between sexes. Similarly, García [52] tested tibial circumference at the nutrient foramen parameter on a Portuguese contemporary collection and a medieval sample reaching different levels of correct classification (78% and 90% respectively). Holland [53] achieved between 85-100% correct sex determination using five measurements from the proximal tibia on an American collection. Kieser et al. [54] also obtained great results using proximal tibia for sexing Caucasoid and Negroid specimens from the Dart Collection. A recent study conducted by Kotěrová et al. [55] used measurements taken on a Czech sample to test the reliability of existing sex estimation discriminant functions. The results showed the decrease in sex estimation accuracy when other methods are applied on their population suggesting the use of population specific formula.

The current study confirms the existence of sexual dimorphism of the tibia that has been reported extensively in the literature [e.g. 49,51]. In addition, the study demonstrates the utility of tibia as a predictor of sex in Greek-Cypriots and Cretans. Discriminant function analysis resulted in multiple univariate and multivariate equations with classification accuracy ranging from 78% to 87% for Greek-Cypriots and from 69-90% for Cretans (see Table 4 and 5). The best single variable for Cretans was LB (82.7%) and for Greek-Cypriots UB (85.2%) similar to other studies [20, 22]. The best sex determination formula for Greek-Cypriots included two variables (MinCirc and LB) reaching 87% of correct sex classification. The accuracy rate is higher for Cretans achieving an accuracy of 89% using three variables (ML, UB and NFap). When the two samples were pooled together accuracy reached 87% using FP2 (ML, NFap and LB) which is similar to the results of other studies with comparable sample sizes (Table 6). FP2 correctly classified about one third of the sample with over 95% confidence. This formula is fairly reliable for both populations and thus it should be used when the ethnicity is not established.

An important finding of this study was the fact that ML was an important sex predictor for Cretans and Greek-Cypriots. ML was selected by the stepwise procedure for all samples (see FCr2 and FCy2, FP2, table 5). This is a consistent finding in all sex estimation studies based on the Cretan sample [4, 5, 7] in contrast with other studies on different European samples [20, 22]. As there are no published studies to date on contemporary Greek-Cypriots, it remains to be seen whether this is going to be a trend for this population as well.

We compared mean differences of the two samples for males and females and found consistent differences for ML and NFap. The proportions of the nutrient foramen were also compared ($\text{NFratio} = 100 \times \text{NFtrsv} / \text{NFap}$) and were found to be statistically different ($p < 0.05$) between

Cretans (Males=70.3%, Females=73%) and Cypriots (Males=67%, Females=69%). In other words, Cretans seem to have more platycnemic tibiae compared to Greek-Cypriots for both sexes. Also, tibial breadth was pointed as a powerful sexual indicator; this parameter is more sexually dimorphic than other areas of the bone due to their functional role in weight loading and muscular stress [56,57]. In addition, the mean maximum length for Cretans is consistently lower compared to Greek-Cypriots (see Table 3). These differences may have an impact in the higher classification accuracy reported for Cretans (Table 5). Alternatively this may simply be a result of the sampling effect.

Cretans are Greeks located in the southernmost island of Greece in the Mediterranean. A comparative study using skulls from the Cretan collection (from which our study sample comes from) and a Minoan sample from Crete reported brachycephalisation of modern Cretans which could be associated with environmental influences and/or gene flow from the East [58]. Genetic evidence, however, supports that modern Cretans from Heraklion prefecture and Lasithi Plateau in Crete are very similar to Minoans [58] which indicates very little admixture with the different invaders in the island for the past 4000 years. In addition, Cretans are genetically further away from mainland Greeks from Attiki, Lakonia, Chios which could explain discrepancies in the expression of sexual dimorphism between the Athens and the Cretan collection as it has been seen in recent comparisons [7, 8].

Cypriots generally consider themselves to be "Greek-blooded", share same language (in a form of a local dialect), dietary habits and religion. A recent genetic study looking at admixture patterns between 96 populations found Greek markers accounting for around 23% of the DNA in Cypriots [59]. Yet, there are Greeks with Cypriot markers barely reaching 12% [60]. The biggest DNA contributors to the Modern Greek genome were Polish 30%, followed by Italians, Iranians, Jordanians and Syrians. Cypriots carried 20% Italian genetic markers and smaller percentages of Iranian, Sicilian, Armenian, Syrian, Georgian, Saudi and Palestinian markers. Hughey et al.'s [59] study placed Cypriots closer to Georgians and Greeks from Chios and Laconia but further away from Cretans (see Figure 5 in Hughey et al. [59]). These data could partly explain why Cypriots are not as close to Cretans as one could expect from cultural and environmental similarities and geographic vicinity.

Nevertheless, the pooled sample gave reasonably high classification accuracy for several formulae which can justify their use in forensic settings. Similarly formulae developed from Greek, Spanish and Italian samples using 3 measurements on the tibia gave very good classification results [21] and can therefore be recommended for Southern Europeans in the absence of population specific standards.

Conclusions:

Both Cretans and Greek-Cypriots populations have exhibited substantial sexual dimorphism of the tibia producing formulae with up to 90% classification accuracy. The pooled sample reached 87% accuracy. We would recommend the application of multivariate formulae suggesting caution on the posterior probability of correct classification of any unknown case. Estimates with over 95% of posterior probabilities can be considered reliable while any less than 80% should be treated with caution. Thus if the bone is complete and the ethnicity is known FCy1 and FCr2 should be used as they correctly classify a larger amount of the sample with over 95% probability. If the exact ethnicity is unknown but one hypothesises that the individual is from a Greek speaking region, the pooled formula FP2 should be used. We hope this work can contribute to the biological profiling and identification of the missing Greek-Cypriots and to potential forensic cases of unknown skeletal remains both in Cyprus and Crete.

Conflict of interest: The authors declare that they have no conflict of interest.

Acknowledgments: The authors would like to thank the Orthodox Church in Limassol (Cyprus) for permitting access to the skeletal material of the ossuary. EFK, JGGD and HL were supported by the Challenge Investment Fund of the University of Edinburgh. PSAP was supported by the CNPq, National Council for Scientific and Technological Development – Brazil. We are grateful to the anonymous reviewers for their contribution in the improvement of our manuscript.

References:

1. D. Ceker, W.D. Stevens, Recovery of Missing Persons in Cyprus: Heavy Equipment Methods and Techniques for Complex Well Excavations, *J. Forensic Sci.* 60 (2015) 1529–1533.
2. <http://www.cmp-cyprus.org/> (accessed 21.02.16)
3. T.W. Phenice, A newly developed visual method of sexing the os pubis, *Am. J. Phys. Anthropol.* 30 (1969) 297-301
4. W.M. Krogman, M.Y. İşcan, *Human Skeleton in Forensic Medicine*, Charles C. Thomas, USA, 1986.
5. S.J. Decker, S.L. Davy-Jow, J.M. Ford, D.R. Hilbelink, Virtual determination of sex: metric and nonmetric traits of the adult pelvis from 3D computed tomography models, *J. Forensic Sci.* 56 (2015) 1107-1114.
6. P.L. Walker, Sexing skulls using discriminant function analysis of visually assessed traits. *Am. J. Phys. Anthropol.* 136 (2008) 39–50
7. C.H. Weiss. *Evaluation Research: methods for assessing program effectiveness*, Englewood Cliffs, New Yersey, 1972.
8. M. Steyn, M.L. Patriquin, Osteometric sex determination from the pelvis-does population specificity matter?, *Forensic Sci. Int.* 191(2009) 113.e1-11.e5.
9. E.F. Kranioti, M.Y. İşcan, M. Michalodimitrakis M, Craniometric analysis of the modern Cretan population, *Forensic Sci. Int.* 180 (2008) 110.e1–110.e5.
10. J.V. Taylor, R. Dibennardo, Determination of sex of White femora by discriminant function analysis: Forensic Implications. *J Forensic Sci*, 27 (1982) 417-423.
11. D.L. France, Observational and metric analysis of sex in the skeleton, in: K.J. Reichs (Ed.), *Forensic osteology: advances in the identification of human remains*, Charles C Thomas, Springfield, USA, 1998, pp. 163-186.
12. M.Y. İşcan, P. Miller-Shaivitz , Sexual dimorphism in the femur and tibia, in: K.J. Reichs, (Ed.), *Forensic osteology: advances in the identification of human remains*, Charles C Thomas, Springfield, USA, 1998, pp. 102-111.
13. M.Y. İşcan, M. Yoshino, S. Kato, Sex determination from the tibia: standards for contemporary Japan, *J. Forensic Sci.* 39 (1994) 785-792
14. M. Steyn, M.Y. İşcan MY, Sex determination from the femur and tibia in South African whites, *Forensic Sci. Int.* 90 (1997) 111-119.
15. M. Šlaus, Z. Tomičić, Discriminant function sexing of fragmentary and complete tibiae from medieval Croatian sites, *Forensic Sci. Int.* 147 (2005) 147-152.
16. E. González-Reimers , J. Velasco-Vázquez , M. Arnay-de-la-Rosa, F. Santolaria-Fernández, Sex determination by discriminant function analysis of the right tibia in the prehispanic

- population of the Canary Islands, *Forensic Sci. Int.* 108 (2000) 165-172.
17. T.D. Holland, Sex assessment using the proximal tibia, *Am. J. Phys. Anthropol.* 85 (1991) 221-227.
 18. J.A. Kieser, J. Moggi-Cecchi, H.T. Groeneveld HT, Sex allocation of skeletal material by analysis of the proximal tibia, *Forensic Sci. Int.* 56 (1992) 29-36.
 19. E. Pomeroy, S.R. Zakrzewski, Sexual dimorphism in diaphyseal cross-sectional shape in the medieval Muslim population of Écija, Spain, and Anglo-Saxon Great Chesterford, UK. *Int. J. Osteoarchaeol.* 19 (2009) 50–65. DOI: 10.1002/oa.981
 20. A. Kotěrová , J. Velemínská, J. Dupej, H. Brzobohatá, A. Pilný, J. Brůžek, Disregarding population specificity: its influence on the sex assessment methods from the tibia, *Int. J. Legal Med.* 2016 (in press).
 21. E.F. Kranioti, M. Apostol, Sexual dimorphism of the tibia in contemporary Greeks, Italians and Spanish: forensic implications, *Int. J. Legal Med.* 36 (2015) doi:10.1007/s00414-014-1045-6. DOI: 10.1007/s00414-014-1045-6.
 22. O. López-Costas, C. Rissech, G. Tranco, D. Turbón, Postnatal ontogenesis of the tibia. Implications for age and sex estimation, *Forensic Sci. Int.* 10 (2012) 207-211.
 23. M.A. Verhoff, F. Ramsthaler, J. Krähahn, U. Deml, R.J. Gille, S. Grabherr , M.J. Thali, K. Kreutz, Digital forensic osteology - possibilities in cooperation with the Virtopsy project, *Forensic Sci. Int.* 174 (2008) 152-156.
 24. S. Grabherr , C. Cooper, S. Ulrich-Bochsler , T. Uldin, S. Ross, L. Oesterhelweg, S. Bolliger, A. Christe, P. Schnyder, P. Mangin, M.J. Thali, Estimation of sex and age of "virtual skeletons"--a feasibility study, *Eur. Radiol.* 19 (2009) 419-29. doi: 10.1007/s00330-008-1155-y.
 25. E. Inci, O. Ekizoglu, R. Turkay, S. Aksoy, I.O. Can IO, D. Solmaz, I. Sayin. Virtual Assessment of Sex: Linear and Angular Traits of the Mandibular Ramus Using Three-Dimensional Computed Tomography, *J. Craniofac. Surg.* 2016 (in press).
 26. J. Singh, R.K. Pathak, Morphometric sexual dimorphism of human sternum in a north Indian autopsy sample: sexing efficacy of different statistical techniques and a comparison with other sexing methods, *Forensic Sci. Int.* 228 (2013) 174.e1-10. doi: 10.1016/j.forsciint.2013.03.020.
 27. H. Brzobohatá, V. Krajíček, Z. Horák, J. Velemínská, Sex Classification Using the Three-Dimensional Tibia Form or Shape Including Population Specificity Approach, *Anthropol Anz.* 71(2014) 219-36.
 28. H. Brzobohatá, V. Krajíček, P. Velemínský, L. Poláček, J. Velemínská, The shape variability of human tibial epiphyses in an early medieval Great Moravian population (9th-10th century AD): a geometric morphometric assessment. *Forensic Sci. Med. Pathol.* 2 (2006) 263-8. doi: 10.1385/FSMP:2:4:263.
 29. D. Navega, R. Vicente, D.N. Vieira, A.H. Ross, E. Cunha, Sex estimation from the tarsal bones in a Portuguese sample: a machine learning approach, *Int. J. Legal Med.* 129 (2015) 651-659. doi: 10.1007/s00414-014-1070-5.
 30. M.F. Darmawan, S.M. Yusuf, M.R. Kadir, H. Haron, Comparison on three classification techniques for sex estimation from the bone length of Asian children below 19 years old:

- an analysis using different group of ages, *Forensic Sci. Int.* 247 (2015) 130.e1-11. doi: 10.1016/j.forsciint.2014.11.007.
31. V.A. Papaioannou, E.F. Kranioti, P. Joveneaux, D. Nathena, M. Michalodimitrakis, Sexual dimorphism of the scapula and the clavicle in a contemporary Greek population: Applications in forensic identification, *Forensic Sci. Int.* 217 (2012) 231.e1–231.e7.
 32. D. Nathena, L. Gambaro, N. Tzanakis, M. Michalodimitrakis, E.F. Kranioti, Sexual dimorphism of the metacarpals in contemporary Cretans: Are there differences with mainland Greeks?, *Forensic Sci. Int.* 257 (2015) 515.e1–515.e8.
 33. E.F. Kranioti, N. Tzanakis, Estimation of Sex from the Upper Limb in Modern Cretans with the Aid of ROC-Analysis: A Technical Report, *Forensic Res. Criminol. Int. J.* 1 (2015) 10.15406/frcij.2015.01.00008.
 34. E.F. Kranioti, M. Michalodimitrakis, Sexual dimorphism of the humerus in contemporary Cretans, *J. Forensic Sci.* 54 (2009) 996-1000.
 35. B. Osipov, K. Harvati, D. Nathena, K. Spanakis, A. Karantanas, E.F. Kranioti, Sexual dimorphism of the bony labyrinth: A new age-independent method, *Am. J. Phys. Anthropol.* 151 (2013) 290–301. doi:10.1002/ajpa.22279
 36. E. Zorba, K. Moraitis, S.K. Manolis, Sexual dimorphism in permanent teeth of modern Greeks, *Forensic Sci. Int.* 210 (2011) 74–81.
 37. S.K. Manolis, C. Eliopoulos, C.G. Koiliou, S.C. Fox, Sex determination using metacarpal biometric data from the Athens Collection, *Forensic. Sci. Int.* 193 (2009) 130.e1-130.e6
 38. M.F. Yavuz, M.Y. İşcan, A.S. Çöloğlu, Age assessment by rib phase analysis in Turks, *Forensic Sci. Int.* 98 (1998) 47–54.
 39. O. Celbis, H. Agritmis, Estimation of stature and determination of sex from radial and ulnar bone lengths in a Turkish corpse sample, *Forensic Sci. Int.* 158 (2006) 135–139.
 40. A. Balseven-Odabasi, E. Yalcinozan, A. Keten, R. Akçan, A.R. Tumer, A. Onan, Age and sex estimation by metric measurements and fusion of hyoid bone in a Turkish population, *J. Forensic Leg. Med.* 20 (2013) 496–501.
 41. M.A. Gudek, A. Uzun, Anthropometric measurements of the orbital contour and canthal distance in young Turkish, *J. Anat. Soc. India* 64 (2015) S1–S6.
 42. S.A. Ulijaszek, D.A. Kerr Anthropometric measurement error and the assessment of nutritional status, *Br. J. Nutr.* 82 (1999) 165–77.
 43. D.G. Altman, J.M. Bland, Statistics notes: the normal distribution. *BMJ* 310 (1995) (6975):298.
 44. A. Ghasemi, S. Zahediasl, Normality Tests for Statistical Analysis: A Guide for Non-Statisticians, *Int. J. Endocrinol. Metab.* 10(2012) 486-489.
 45. Scientific Working Group for Forensic Anthropology (2010) Sex assessment. Issued

- March 6th, 2010, from <http://swganth.org/products-drafts.html>. (accessed 03.03.16)
46. Code of Practise. British Association for Forensic Anthropology.
<http://www.bahid.org/bafa/cpd>. (accessed 04.04.16)
 47. FASE Basic Workshop in Forensic Anthropology, 1-5 September, 2015
<http://lfa.uc.pt/event/fase-basic-workshop-in-forensic-anthropology> (accessed 15.08.16)
 48. M.Y.Isçan, Forensic Anthropology of sex and body size, *Forensic Sci. Int.* 147 (2005) 107-112.
 49. M. Y. Slaus, Z. Tomicic, Discriminant function sexing of fragmentary and complete tibiae from medieval Croatians sites, *Forensic Sci. Int.* 147 (2005) 147-152.
 50. M.Y. Isçan, P. Miller-Shaivitz, Determination of sex from the tibia, *Am. J. Phys. Anthropol.* 64 (1984) 53-57.
 51. E. González-Reimers, J. Velasco-Vázquez, M. Arnay-de-la-Rosa, F. Santolaria-Fernández, Sex determination by discriminant function analysis of the right tibia in the prehispanic population of the Canary Islands, *Forensic Sci. Int.* 108 (2000) 165-172.
 52. S. Garcia, Is the circumference at the nutrient foramen of the tibia of value to Sex determination on Human Osteological Collections? Testing a new method, *Int. J. Osteoarchaeol.* 22 (2000) 361-365.
 53. T.D. Holland, Sex assessment using the proximal tibia, *Am. J. Phys. Anthropol.* 85 (1991) 221-227.
 54. J.A. Kieser, J. Moggi-Cecchi, H.T. Groeneveld, Sex allocation of skeletal material by analysis of the proximal tibia, *Forensic Sci. Int.* 56 (1992) 29-36.
 55. A. Kotěrová, J. Velemínská, J. Dupej, H. Brzobohatá, A. Pilný, J. Brůžek, Disregarding population specificity: its influence on the sex assessment methods from the tibia, *Int. J. Legal Med.* (2016) DOI: 10.1007/s00414-016-1413-5
 56. T.K. Black, A new method for assessing the sex of fragmentary skeletal remains: Femoral Shaft Circumference, *Am. J. Phys. Anthropol.* 48 (1978) 227-232.
 57. J.V. Taylor, R. Dibennardo, R. . Determination of sex of White femora by discriminant function analysis: Forensic Implications, *J. Forensic Sci.* 27 (1982) 417-423.
 58. E. Kranioti, Craniometric Analysis of the Cretan population, «Εν Γορτύνη και Αρκαδία εγένετο», Messara, Crete, Greece. In P. E., & D. M. (Eds.), *Proceedings of the 1st International Multidisciplinary Conference on the history and civilisation of Southern Crete «Εν Γορτύνη και Αρκαδία εγένετο»*. Heraklion, 2014: 615-623.
 59. J.R. Hughey, P. Paschou, P. Drineas, D. Mastropaolo, D.M. Lotakis, P.A. Navas, M.

- Michalodimitrakis, J.A. Stamatoyannopoulos, G. Stamatoyannopoulos, A European population in Minoan Bronze Age Crete, *Nat. Commun.* 4 (2013):1861
doi:10.1038/ncomms2871.
60. G. Hellenthal, G.B.J. Busby, G. Band, J.F. Wilson, C. Capelli, D. Falush, S. Myers, A. Genetic Atlas of Human Admixture History, *Science* 348 (2014) 747-751.
doi:10.1126/science.1243518.

Table 1. Intra- and Inter-observer error is quantified by calculating TEM, rTEM and R for each variable.

	Intra-Observer Error (N=30)			Inter-Observer Error (N=15)		
	TEM	rTEM	R	TEM	rTEM	R
TL	0.56	0.16	0.99	1.88	0.51	0.96
UB	0.45	0.66	0.99	1.3	1.89	0.70
NFap	0.59	1.79	0.95	0.35	1.02	0.87
NFtrsv	0.43	1.90	0.94	0.6	2.3	0.69
NFCirc	0.73	0.87	0.99	2.4	2.75	0.79
NFmin	0.70	0.98	0.98	1	1.36	0.85
LB	0.74	1.50	0.94	0.95	1.93	0.86

Table 2. Results for mean differences between males and females for all variables in both samples.

Greek-Cypriots								
Males				Females				
	N	Mean	SD	N	Mean	SD	F-value	P-Value
ML	67	374.18	21.28	58	341.57	21.65	71.82	0.001
UB	63	74.17	4.07	59	67.59	5.02	63.6	0.001
NFap	70	35.85	2.86	62	30.69	3.51	86.13	0.001
NFtrsv	70	23.92	2.11	62	21.13	2.50	48.47	0.001
NFCirc	70	94.57	6.47	62	82.06	8.43	92.64	0.001
MinCirc	70	75.74	5.43	62	67.06	5.79	79.03	0.001
LB	66	44.70	2.92	58	39.68	3.01	88.39	0.001
Cretans								
Males				Females				
	N	Mean	SD	N	Mean	SD	F-value	0.001
ML	85	362.75	19.55	72	332.24	17.71	103.48	0.001
UB	85	75.29	3.90	72	68.47	3.96	117.39	0.001
NFap	85	35.17	2.36	72	30.86	2.37	130.05	0.001
NFtrsv	85	24.69	2.22	72	22.33	1.99	48.583	0.001
NFCirc	85	94.37	6.44	72	84.06	5.90	107.98	0.001
Mincirc	85	74.45	4.75	72	68.25	4.50	69.67	0.001
LB	85	45.02	2.78	71	40.47	2.60	110.15	0.001

Table 3. Independent T-test of mean differences between Greek-Cypriots and Cretans for each subgroup. Bold values indicate statistically significant differences.

		Male					Female				
		N	Mean	SD	t-value	p-value	N	Mean	SD	t-value	p-value
ML	Cretans	85	362.75	19.55	-3.440	<0.05	72	332.24	17.71	-2.704	<0.05
	Cypriots	67	374.18	21.28			58	341.57	21.65		
UB	Cretans	85	75.29	3.90	1.695	NS	72	68.47	3.96	1.120	NS
	Cypriots	63	74.17	4.07			59	67.59	5.02		
NFap	Cretans	85	35.17	2.36	-1.614	NS	72	30.86	2.37	0.312	NS
	Cypriots	70	35.84	2.86			62	30.69	3.51		
NFtrsv	Cretans	85	24.69	2.22	2.211	<0.05	72	22.33	1.99	3.099	<0.05
	Cypriots	70	23.92	2.11			62	21.13	2.50		
NFCirc	Cretans	85	94.37	6.44	-0.196	NS	72	84.06	5.90	1.560	NS
	Cypriots	70	94.57	6.47			62	82.06	8.42		
MinCirc	Cretans	85	74.45	4.75	1.333	NS	72	68.25	4.50	1.333	NS
	Cypriots	70	75.74	5.43			62	67.06	5.79		
LB	Cretans	85	45.02	2.78	0.700	NS	71	40.47	2.60	1.589	NS
	Cypriots	66	44.70	2.92			58	39.68	3.01		

NS=Non-significant ($p>0.05$)

Table 4. Demarking points for univariate function and classification accuracies for original and cross-validated samples.

	Greek-Cypriots	Original					Cross-validated				
		Male		Female		Total	Male		Female		Total
	Demarking	N	%	N	%	%	N	%	N	%	%
TL	357.9	56/67	83.6	46/58	79.3	81.6	56/67	83.6	46/58	79.3	81.6
UB	70.8	55/63	87.3	49/59	83.1	85.2	55/63	87.3	49/59	83.1	85.2
NFap	33.3	58/70	82.9	50/52	80.6	81.8	58/70	82.9	50/62	80.6	81.8
NFtrsv	22.5	52/70	74.3	51/62	82.3	78	52/70	74.3	51/62	82.3	78
NFCirc	88.3	61/70	87.1	51/62	82.3	84.8	61/70	87.1	51/62	82.3	84.8
MinCirc	71.4	57/70	81.4	50/62	80.6	81.1	57/70	81.4	50/62	80.6	81.1
LB	42.2	53/66	80.3	48/58	82.8	81.5	53/66	80.3	48/58	82.8	81.5
	Cretans	Original					Cross-validated				
		Male		Female		Total	Male		Female		Total
	Demarking	N	%	N	%	%	N	%	N	%	%
TL	347.5	66/85	77.6	59/72	81.9	79.6	66/85	77.6	59/72	81.9	79.6
UB	71.9	69/85	81.2	58/72	80.6	80.9	69/85	81.2	58/72	80.6	80.9
NFap	33	68/85	80	59/72	81.9	80.9	68/85	80	58/72	80.6	80.3
NFtrsv	23.5	57/85	67.1	52/72	72.2	69.4	57/85	67.1	52/72	72.2	69.4
NFCirc	89.2	66/85	77.6	60/72	83.3	80.3	66/85	77.6	60/72	83.3	80.3
MinCirc	71.3	59/85	69.4	58/72	80.6	74.5	59/85	69.4	58/72	80.6	74.5
LB	42.5	68/85	80	60/71	84.5	82.1	68/85	80	60/71	84.5	82.1
	Pooled sample	Original					Cross-validated				
		Male		Female		Total	Male		Female		Total
	Demarking	N	%	N	%	%	N	%	N	%	%
TL	352.1	118/151	77.6	109/130	83.8	80.5	118/151	77.6	109/130	83.8	80.5
UB	71.4	126/148	85.1	107/131	81.7	83.5	126/148	85.1	107/131	81.7	83.5
NFap	33.1	126/155	81.3	110/134	82.1	81.7	126/155	81.3	110/134	82.1	81.7
NFtrsv	23.1	110/155	71	102/134	76.1	73.4	110/155	71	102/134	76.1	73.4
NFCirc	88.8	129/155	83.2	107/134	79.9	81.7	129/155	83.2	107/134	79.9	81.7
MinCirc	71.4	116/155	74.8	108/134	80.6	77.5	116/155	74.8	108/134	80.6	77.5
LB	42.4	119/151	78.8	110/129	85.3	81.8	119/151	78.8	110/129	85.3	81.8

Table 5. Multivariate discriminant functions for Cretans and Greek-Cypriots, classification accuracy for original and cross-validation data and percentage of accuracy with posterior probability (PP) >95%. Sectioning point is set to zero in all cases.

									Original					Cross-validated					
									Male		Female		To tal	Male		Female		To tal	PP> 95 %
Func tions	M L	U B	Nf ap	NFt rsv	NF Cir c	Min Circ	LB	Con stan t	N	%	N	%	%	N	%	N	%	%	%
FCy1	0.015	0.014	0.063	- 0.134	0.029	0.016	0.193	- 17.197	52/60	86.7	45/53	84.9	85.8	52/60	86.7	45/53	84.9	85.8	29.2
FCy2	0.021						0.243	- 17.868	57/66	86.4	51/58	87.9	87.1	55/66	83.3	51/58	87.9	85.5	27.4
FCy3		0.1	0.091	- 0.002	0.048			- 14.286	57/63	90.5	46/58	79.3	85.1	56/63	88.9	46/58	79.3	84.3	16.5
FCy4						0.085	0.218	- 15.238	58/66	87.9	49/57	86	87.0	58/66	87.9	49/57	86.0	87.0	16.5
FCy5			0.099	- 0.026	0.101			- 11.62	63/70	90	49/62	79	84.8	63/70	90.0	49/62	79	84.8	18.2
FCr1	0.019	0.069	0.208	- 0.091	0.019	- 0.009	0.099	- 21.568	79/85	92.9	62/71	87.3	90.4	79/85	92.9	61/71	85.9	89.7	42.4
FCr2	0.021	0.09	0.231					- 21.525	79/85	92.9	62/72	86.1	89.8	79/85	92.9	62/72	86.1	89.8	44.6
FCr3		0.15	0.243	- 0.103	0.031			- 19.041	77/85	90.6	63/72	87.5	89.2	75/85	88.2	62/72	86.1	87.3	41.4
FCr4						0.068	0.291	- 17.305	68/85	80	60/71	84.5	82.1	68/85	80.0	60/71	84.5	82.1	21.2
FCr5			0.312		0.05			- 14.749	69/85	81.2	60/72	83.3	82.2	69/85	81.2	60/72	83.3	82.2	29.3
FP1	0.014	0.049	0.161	- 0.044	0.005	- 0.016	0.149	- 18.236	126/144	87.5	106/125	84.8	86.2	125/144	86.8	103/125	82.4	84.8	31.0
FP2	0.016		0.161				0.164	- 17.85	135/150	90	109/129	84.5	87.5	134/150	89.3	109/129	84.5	87.1	31.2
FP3		0.123	0.179	- 0.003	0.023			- 16.054	132/147	89.8	108/131	82.4	86.3	131/147	89.1	107/131	81.7	86.6	19.8
FP4						0.075	0.246	- 15.829	123/150	82	109/129	84.5	83.2	122/150	81.3	109/129	84.5	82.8	18.3
FP5			0.211		0.064			- 12.683	130/154	84.4	109/135	80.7	82.7	130/154	84.4	109/135	80.7	82.7	17.8

Table 6. Metric studies on sexual dimorphism using the tibia.

Author/Year	Nº of variables	Population	Accuracy rates
Holland (1991)	1 variable	Hamann-Todd Collection (N=100)	86–95%
Slaus and Tomicic (2005)	6 variables	medieval Croatian population (N=180)	81.7-92.2%
Slauc et al. (2013)	6 variables	modern Croatian population (N=180)	84.4-91.1%
Isçan and Miller-Shaivitz (1984)	4 variables	Terry Collection (N=159)	80-84%
Gonzalez-Reimersa et al. (2000)	7 variables	Canary Island population (N=59)	94.9-98.3%
García (2010)	1 variable	Portugal (N=57)	78-90%
Kieser et al. (1992)	5 variables	Dart Collection (N=202)	84.62–92%
Kotěrová et al. (2016)	10 variables	Czech sample (N=56)	82.1-87.5 %
Our study	7 variables	Cretans (N=157) and Greek-Cypriots (N=132)	78-90%